PAIN® 154 (2013) S87–S93

PAIN®

www.elsevier.com/locate/pain

Review

# Endocannabinoids: A unique opportunity to develop multitarget analgesics

Sabatino Maione [a], Barbara Costa [b], Vincenzo Di Marzo [c,*]

[a] Endocannabinoid Research Group, Department of Experimental Medicine – Division of Pharmacology 'L. Donatelli', Second University of Naples, Naples, Italy
[b] Department of Biotechnology and Bioscience, University of Milano-Bicocca, Milan, Italy
[c] Endocannabinoid Research Group, Institute of Biomolecular Chemistry – C.N.R., Pozzuoli, Italy

*Sponsorships or competing interests that may be relevant to content are disclosed at the end of this article.*

ARTICLE INFO

*Article history:*
Received 8 August 2012
Received in revised form 3 January 2013
Accepted 12 March 2013

*Keywords:*
Biennial review of pain 2013
Pain
Endocannabinoids

ABSTRACT

After 4 millennia of more or less documented history of cannabis use, the identification of cannabinoids, and of $\Delta^9$-tetrahydrocannabinol in particular, occurred only during the early 1960s, and the cloning of cannabinoid $CB_1$ and $CB_2$ receptors, as well as the discovery of endocannabinoids and their metabolic enzymes, in the 1990s. Despite this initial relatively slow progress of cannabinoid research, the turn of the century marked an incredible acceleration in discoveries on the "endocannabinoid signaling system," its role in physiological and pathological conditions, and pain in particular, its pharmacological targeting with selective agonists, antagonists, and inhibitors of metabolism, and its previously unsuspected complexity. The way researchers look at this system has thus rapidly evolved towards the idea of the "endocannabinoidome," that is, a complex system including also several endocannabinoid-like mediators and their often redundant metabolic enzymes and "promiscuous" molecular targets. These peculiar complications of endocannabinoid signaling have not discouraged efforts aiming at its pharmacological manipulation, which, nevertheless, now seems to require the development of multitarget drugs, or the re-visitation of naturally occurring compounds with more than one mechanism of action. In fact, these molecules, as compared to "magic bullets," seem to offer the advantage of modulating the "endocannabinoidome" in a safer and more therapeutically efficacious way. This approach has provided so far promising preclinical results potentially useful for the future efficacious and safe treatment of chronic pain and inflammation.



## 1. A rapid excursus from ancient medicinal cannabis to the modern "endocannabinoidome"

The medicinal use of cannabis against pain seems to date back several millennia, and probably originates in ancient China. Shen Nung (the "red emperor," 2838–2698 BC), considered the father of all herbalists, is said to have suggested its use in his book "The Herbal" (of which, however, only much more recent copies exist). However, the earliest written reference to the use of hemp against pain and inflammation is the Chinese Rh–Ya (~1500 BC). Ever since, evidence exists for the use of cannabis against various inflammatory and painful conditions in the ancient Egyptian, Indian, Greek, and Roman pharmacopeias, and also in medieval Islamic medicine. More recently, the Irish physician William O'Shaughnessy is credited with introducing the therapeutic use of cannabis to Western medicine in the 1830s, and Queen Victoria's personal physician, Sir Robert Russell, is said to have suggested its use against menstrual pains and cramps [59].

It took, then, more than one century to identify, in cannabis flowers, the chemical components likely to be responsible for these therapeutic actions: a series of terpenophenolic natural products known as "cannabinoids" (Fig. 1). Cannabidiol (CBD) and $\Delta^9$-tetrahydrocannabinol (THC), both present in the plant as their acid derivatives and produced after the heating or drying of the flowers, were the first such compounds to be fully characterized in the early 1960s, and it was soon clear that the latter, and not the former, was responsible for the psychotropic effects of cannabis and its recreational preparations [33,54]. This probably explains why, for several decades, research on CBD lagged behind, and most experimental efforts were concentrated to understand the mechanism of action of THC, possibly to help prevent what, in the 1960s and 1970s, seemed to be the dangerous consequences of marijuana abuse. These efforts culminated, although only between the late 1980s and early 1990s, with the identification of 2 G protein-coupled receptors (GPCRs) for THC, named cannabinoid receptor type-1 ($CB_1$) and type-2 ($CB_2$) [52,57].

The existence of GPCRs for a xenobiotic compound clearly suggested the presence, in animal tissues expressing such receptors,

* Corresponding author. Address: Endocannabinoid Research Group, Institute of Biomolecular Chemistry – C.N.R., Via Campi Flegrei 34, Comprensorio Olivetti, 80078 Pozzuoli (NA), Italy. Tel.: +39 081 8675093; fax: +39 081 8041770.
*E-mail address:* vdimarzo@icb.cnr.it (V. Di Marzo).

0304-3959/$36.00 
http://dx.doi.org/10.1016/j.pain.2013.03.023

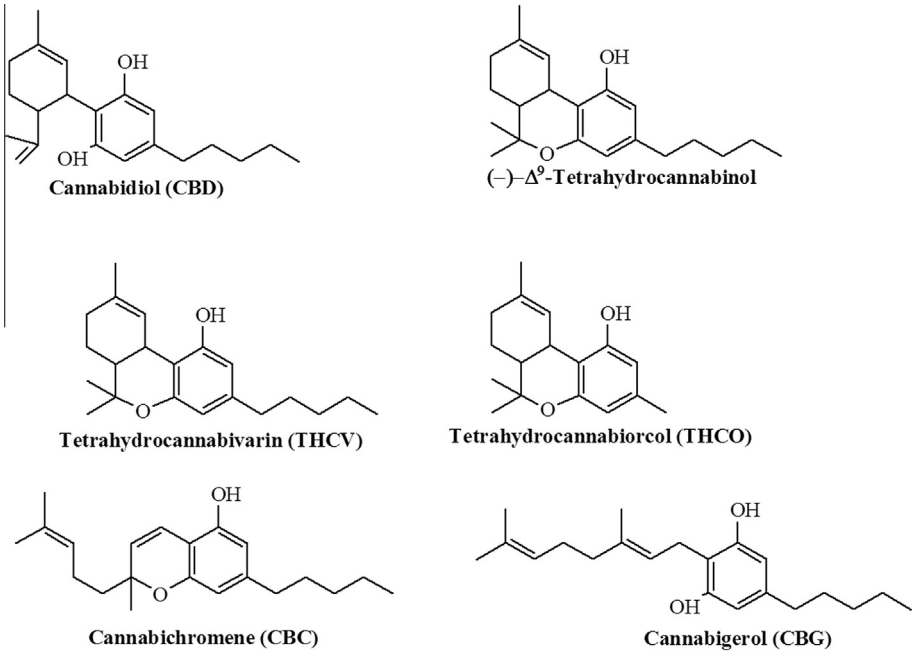


**Fig. 1.** Chemical structures of the most studied cannabinoids from *Cannabis sativa*.

of endogenous ligands, which were indeed identified in the 1990s as 2 arachidonic acid-derived mediators, anandamide (*N*-arachidonoyl-ethanolamine [AEA]) and 2-arachidonoylglycerol (2-AG) [22,53,73], baptized in 1995 as "endocannabinoids" [26]. These discoveries, in turn, literally unchained a rapid series of events, including: (1) the finding of metabolic (both anabolic and catabolic) routes and corresponding enzymes for the separate regulation of AEA and 2-AG levels [23]; (2) the development of methods for endocannabinoid analysis in tissues and biological fluids, and the understanding that endocannabinoid concentrations are altered during certain pathological states selectively in tissues affected by those conditions [47]; (3) the increasing realization that endocannabinoids not only are rather promiscuous mediators and interact at physiopathological concentrations with other receptors (be they channels, nuclear receptors or non-$CB_1$, non-$CB_2$ GPCRs) [20], but are also biosynthesized and degraded through seemingly redundant metabolic pathways and enzymes; and (4) the identification of endocannabinoid-like bioactive fatty acid amides, often accompanying AEA in tissues and sharing some of its biosynthetic or inactivating pathways, but usually acting at non-$CB_1$, non-$CB_2$ receptors [2]. As a consequence of this series of discoveries, the initial concept of "endocannabinoid signaling system" as the ensemble of endocannabinoids, their metabolic enzymes, and $CB_1$ and $CB_2$ receptors, evolved into the as-yet not fully developed idea of the "endocannabinoidome," that is, a more complex system including endocannabinoids and endocannabinoid-like mediators (ELMs) and their often redundant metabolic enzymes and "promiscuous" molecular targets (Table 1). It is clear that, in view of these recent developments, the pharmacological manipulation of endocannabinoid levels and action for therapeutic purposes, initially seen as a safer alternative to the direct targeting of psychotropic $CB_1$ and immunomodulatory $CB_2$ receptors [25], poses new challenges to medicinal chemists, pharmacologists, and clinicians. Nevertheless, as will be discussed in this article, in view of the new technologies provided by the explosion of the "omic" era, and of the understanding that a more holistic approach is needed to study pain and inflammation (as with other pathologies), the complexity of the "endocannabinoidome" also offers unprecedented opportunities for the development of novel "multitarget" analgesic drugs, and for the revisitation of "old" medicines and natural products.

## 2. Endocannabinoid-based drugs and "old" medicines: from "magic bullets" to multitarget drugs

Given the widely accepted concept that endocannabinoids are "pro-homeostatic" mediators produced "on demand" to act specifically at the site of pathological conditions [23,47], it was reasoned that drugs inhibiting their inactivation would prolong the effects of $CB_1$ and $CB_2$ action in a more tissue-specific manner than "direct" $CB_1$ and $CB_2$ agonists. Therefore, considerable effort has been devoted in recent years to developing selective inhibitors of fatty acid amide hydrolase-1 (FAAH-1) and monoacylglycerol lipase (MAGL), which are responsible for most of AEA and 2-AG enzymatic hydrolysis, respectively [25] (Table 1). These compounds have provided very promising results in experimental models of chronic and inflammatory pain and inflammation [32]. However, there are complications when one inhibits these enzymes. FAAH-1 is also responsible for the hydrolysis of several ELMs acting at non-$CB_1$, non-$CB_2$ molecular targets, some of which, like the transient receptor potential vanilloid type-1 (TRPV1) channel, are also activated by AEA and participate in nociception [24]. Thus, FAAH-1 inhibitors may cause the indirect activation also of such targets, which may interfere with the expected analgesic action of these compounds (although both counteraction and potentiation may occur). On the other hand, MAGL also participates in the generation of arachidonic acid as the biosynthetic precursor for eicosanoids [58]. Whilst some of the latter compounds, such as, for example, prostaglandins $E_2$ and $F_{2\alpha}$, can be proinflammatory and pronociceptive, others (e.g., prostacyclin $I_2$) play important physiological functions. Thus, also inhibiting MAGL may produce adverse events. Furthermore, chronic administration of fully effective doses of MAGL inhibitors causes desensitization of $CB_1$ receptors, and hence tolerance [70], a phenomenon that might be avoided using lower but possibly less effective dosages. Finally, both AEA and, particularly, 2-AG, are inactivated also by other enzymes, for example, some serine hydrolases (FAAH-2 in the case of the former compound, and α,β-hydrolase-6 and -12 and FAAH-1 in the case of 2-AG) [23], the function of which in humans is not known and might include a compensatory role for FAAH-1 and MAGL inactivation. Perhaps more importantly, recent evidence [34,38], based on early in vitro data (reviewed in [66]), indicates that both AEA and

**Table 1**
The "endocannabinoidome": a partial view of endocannabinoids and chemically similar mediators playing a role in pain or inflammation.

| Mediator | Molecular target(s) | Biosynthetic enzyme(s) | Catabolic mechanism(s) |
|---|---|---|---|
| 2-Arachidonoyl-glycerol (2-AG) | • *$CB_1$ = $CB_2$ (agonist)*<br>• $GABA_A$ (allosteric enhancer) | • *DAGLα*<br>• DAGLβ | • *Putative membrane transporter*<br>• MAGL<br>• ABHD6<br>• ABHD12<br>• *FAAH-1*<br>• *COX-2*<br>• 12/*15*-lipoxygenases |
| Anandamide (*N*-arachidonoyl-ethanolamine) | • *$CB_1$ > $CB_2$ (partial agonist)*<br>• *TRPV1 (agonist)*<br>• *TRPM8 (inhibitor)*<br>• *T-type $Ca^{2+}$ channels (inhibitor)*<br>• TASK $K^+$ channels (inhibitor)<br>• *PPAR-γ (agonist)* | • NAPE-PLD<br>• GDE1<br>• ABHD4 | • *Putative membrane transporter*<br>• *FAAH-1*<br>• *COX-2*<br>• 12/*15*-lipoxygenases<br>• *Cytochrome p450* |
| *N*-oleoyl-ethanolamine (OEA) | • *PPAR-α (agonist)*<br>• *TRPV1 (agonist)*<br>• GPR119 (agonist) | • NAPE-PLD<br>• GDE1<br>• ABHD4 | • *FAAH-1*<br>• FAAH-2 |
| *N*-palmitoyl-ethanolamine (PEA) | • *PPAR-α (agonist)*<br>• *TRPV1 (allosteric enhancer)*<br>• *GPR55 (agonist?)*<br>• *PPAR-γ (agonist?)* | • NAPE-PLD<br>• GDE1<br>• ABHD4 | • NAAA<br>• *FAAH-1*<br>• FAAH-2 |
| *N*-arachidonoyl-glycine | • *GPR18 (agonist)*<br>• *T-type $Ca^{2+}$ channels (inhibitor)*<br>• glycine transporter-2 (inhibitor) | • *FAAH-1 (?)* | • *FAAH-1*<br>• 12/*15*-lipoxygenases |
| *N*-acyl-dopamines | • *TRPV1 (agonists, unsaturated ones)*<br>• *$CB_1$ (agonist, arachidonoyl derivative)*<br>• *T-type $Ca^{2+}$ channels (inhibitor)* | • *FAAH-1 (?)* | • *Putative membrane transporter*<br>• COMT |
| *N*-acyl-serotonins | • *TRPV1 (antagonists, unsaturated ones)*<br>• *FAAH-1 (inhibitors, unsaturated ones)* | • Unknown | • Unknown |
| *N*-arachidonoyl-serine | • *GPR55 (agonist?)* | • Unknown | • Unknown |
| Prostamide $F_{2\alpha}$ | • Heterodimer between the FP receptor and a splicing variant thereof (agonist) | • *COX-2*<br>• Prostaglandin F synthase | • Unknown |
| Prostaglandin $E_2$ glycerol ester | • Unknown GPCR (agonist) | • *COX-2*<br>• Prostaglandin E synthase | • MAGL |

ABHD, α,β-hydrolase; COMT, catecholamine *O*-methyl transferase; COX-2, cycloxygenase-2; DAGL, diacylglycerol lipase; FAAH, fatty acid amide hydrolase; GDE1, glycerophosphodiesterase-1; GPCR, G protein-coupled receptor; GPR, orphan GPCR; MAGL, monoacylglycerol lipase; NAAA, *N*-acylethanolamine-hydrolyzing acid amidase; NAPE-PLD, *N*-acyl-phosphatidylethanolamine-specific phospholipase D; PPAR, peroxisome proliferator-activated receptor; TRPM8, transient receptor potential melastatin type-8 cation channel; TRPV1, transient receptor potential vanilloid type-1 cation channel.
*Note.* The proposed molecular targets and biosynthetic and degrading enzymes for each compound or class of mediators are shown in order of importance, determined by the number of independent studies confirming their role in the endocannabinoidome. Question marks refer to data not yet confirmed by 2 or more independent studies. Proteins and mechanisms shown in *italics* can be modulated also by some phytocannabinoids.

2-AG are metabolized by cycloxygenase-2 (COX-2) and, subsequently, prostaglandin synthases to compounds that are now considered part of the "endocannabinoidome," and are known as prostaglandin-ethanolamides (prostamides) and prostaglandin-glycerol esters, respectively (Table 1). These compounds usually exert pronociceptive and proinflammatory effects via nonprostanoid and non-$CB_1$, non-$CB_2$ receptors. Thus, inhibiting FAAH-1 and MAGL might cause endocannabinoids to be processed through these mechanisms instead, thereby counteracting the indirect activation of cannabinoid receptors [24,60].

Thus, many reasons may exist to justify the, perhaps pessimistic, prediction that selective inhibitors of endocannabinoid enzymatic hydrolysis may not necessarily be clinically effective even when devoid of adverse side effects. Indeed, a recent phase II clinical trial with the irreversible FAAH-1 inhibitor, PF-04457845 (Fig. 2), showed that chronic administration of this compound to patients with pain due to osteoarthritis of the knee, whilst well tolerated and efficacious at elevating the plasma levels of the endogenous substrates of the enzyme (i.e., AEA and other *N*-acylethanolamines), failed to produce analgesia [41]. Although there are always several possible reasons for the failure in the clinic of any type of preclinically effective medication, this recent study should perhaps lead to re-evaluation of the possibility of co-administering inhibitors of endocannabinoid inactivation with other analgesics. Based on the above reasoning, one may envisage the co-administration of PF-04457845 with submaximal, and possibly safer, doses of COX-2 inhibitors. Interestingly, evidence (reviewed in [60]) exists to suggest that, at least in animal models, 2 widely used analgesic and antipyretic drugs, acetaminophen (paracetamol) and dipyrone (metamizol), act not just by inhibiting cyclooxygenases per se but also through their metabolites formed in vivo through the action of FAAH-1. These metabolites can then inhibit endocannabinoid inactivation and/or directly activate cannabinoid receptors and/or desensitize or antagonize TRPV1 channels. Therefore, simultaneous modulation of several targets is perhaps the key to efficacious and well-established antiinflammatory therapies, as it might be already enacted by 2 widely used analgesics.

Also, the selective targeting of TRPV1, possibly the therapeutic strategy towards new antihyperalgesic drugs in which the pharmaceutical industry has invested more money in the last decade, has been so far unsuccessful in the clinic, although seemingly for reasons of safety rather than efficacy. In fact, most of the TRPV1 antagonists tested thus far produce worrisome hyperthermic effects at effective doses [75]. On the other hand, activation and desensitization of TRPV1 with topical creams and patches containing capsaicin [10], accompanied by transient peripheral denervation when high concentrations of this compound are used, is also effective against neuropathic pain, although not devoid of side effects [75]. Our

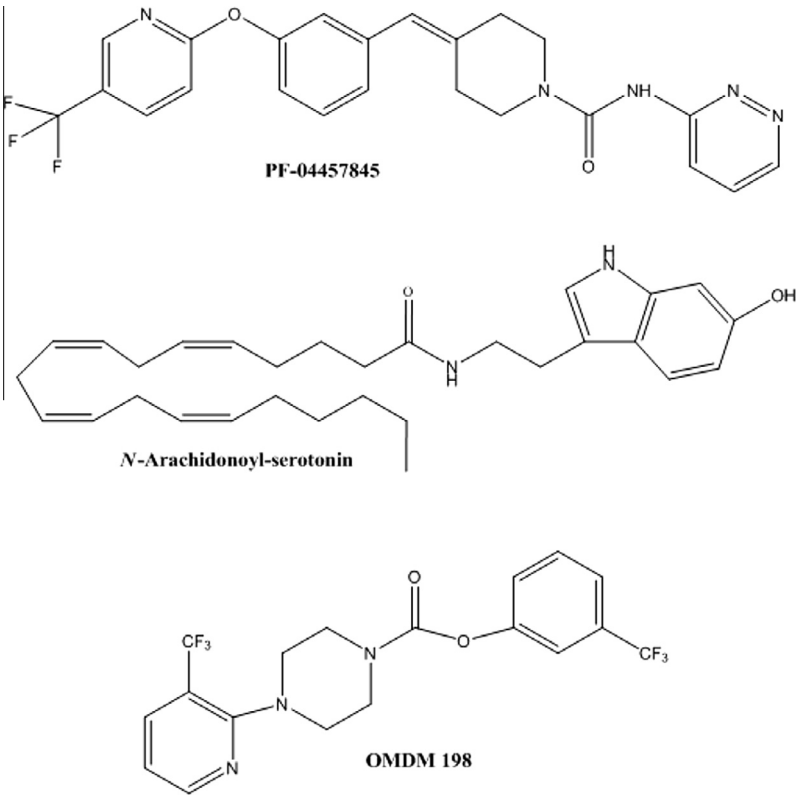


**Fig. 2.** Chemical structures of a selective FAAH-1 inhibitor already tested in the clinic, PF-04457845, and of 2 "dual" FAAH-1/TRPV1 blockers, *N*-arachidonoyl-serotonin and OMDM-198.

groups have recently dedicated some efforts to assessing whether, instead, simultaneous blockade of TRPV1 and FAAH-1 (2 proteins often co-localized in both central and peripheral nervous systems) with the same compound could provide a better strategy. The first dual TRPV1/FAAH-1 blocker ever identified is *N*-arachidonoyl-serotonin [49] (Fig. 2), which was later identified as a naturally occurring ELM [78]. This compound is more efficacious against neuropathic [37], inflammatory [13], and acute [49] pain than singularly administered, more potent FAAH-1 and TRPV1 antagonists. Importantly, *N*-arachidonoyl-serotonin appears to be devoid of hyperthermic effects in mice [13]. In view of the consideration that a certain degree of anxiety in osteoarthritic patients might render antihyperalgesic/antiinflammatory drugs more efficacious [41], it is interesting to note how this compound also proved to be a more potent anxiolytic agent as compared to synthetic drugs with only FAAH-1 or TRPV1 antagonist properties [55], an effect that might represent a plus-value when assessing pain scores in man. Finally, *N*-arachidonoyl-serotonin was recently found to inhibit also another target for analgesic drugs, the T-type $Ca^{2+}$ channels [36].

We have recently managed to improve, if not the pharmacodynamic, at least possibly the stability and bioavailability of *N*-arachidonoyl-serotonin (which contains 2 chemical moieties, the arachidonate and serotonin groups, highly liable to oxidation) with the development of OMDM198 [56] (Fig. 2), a fully synthetic piperazinyl carbamate TRPV1/FAAH-1 blocker. This compound proved effective in models of inflammatory pain and osteoarthritis (our own unpublished data and Starowicz et al., 22nd Symposium on Cannabinoids, Freiburg, Germany, July 22–27, 2012). These preliminary data, although still far from being translatable into suitable clinical treatments, provide at least the proof-of-concept status to the idea that rationally designed, multitarget drugs based on the "endocannabinoidome" may afford more potent, and yet possibly safer, antihyperalgesic and antiinflammatory drugs, and will hopefully foster more intensive studies in this direction.

## 3. Emerging ELM-based drugs

Apart from *N*-arachidonoyl-serotonin, several other endogenous small-molecule components of the "endocannabinoidome" exhibit pain modulatory effects via several mechanisms of action (Fig. 3). Among the best studied ones are: (1) *N*-arachidonoyl-glycine, which inhibits pain [39,79] and has so far been suggested to inhibit T-type $Ca^{2+}$ channels [3] and the glycine transporter-2 [27] and to activate the orphan GPCR, GPR18, also possibly involved in inflammation [44]; and (2) *N*-arachidonoyl-dopamine, a dual TRPV1 and $CB_1$ agonist initially described to induce pain [40] and recently suggested to also inhibit T-type $Ca^{2+}$ channels [64] and to counteract pain when spinally injected via TRPV1 desensitization and $CB_1$ activation [31]. However, among ELMs, the one that has certainly been most thoroughly studied is *N*-palmitoyl-ethanolamine (PEA) (Fig. 3).

PEA is currently marketed in Italy as Normast® (Epitech Group, Saccolongo, Padua, Italy), a dietary supplement with specific medical use, prescribed for the treatment of neuropathic pain and other conditions characterized, among other things, by mast cell hyperactivity. PEA is also marketed as Pelvilen® (Epitech Group, Saccolongo, Padua, Italy) in a 10:1 formulation with the antioxidant agent polydatin, against pelvic pain. PEA and other saturated *N*-acylethanolamines that are almost ubiquitously found in animal tissues, were initially described as autacoid local inflammation antagonist amides, with specific inhibitory action on mast cell degranulation [30]. However, since then, the potential therapeutic effects of this pleiotropic compound have greatly enlarged, encompassing antiinflammatory, analgesic, and neuroprotective actions exerted also at the level of microglia, astrocytes, neurons, macrophages, and adipocytes (see [28] for a recent review). Perhaps more relevant to the subject of this article, PEA is now recognized as a multitarget lipid mediator, its effects being exerted essentially through 3 general mechanisms: (1) activation of peroxisome proliferator-activated receptor-α (PPAR-α), with subsequent short-term (nonnuclear)

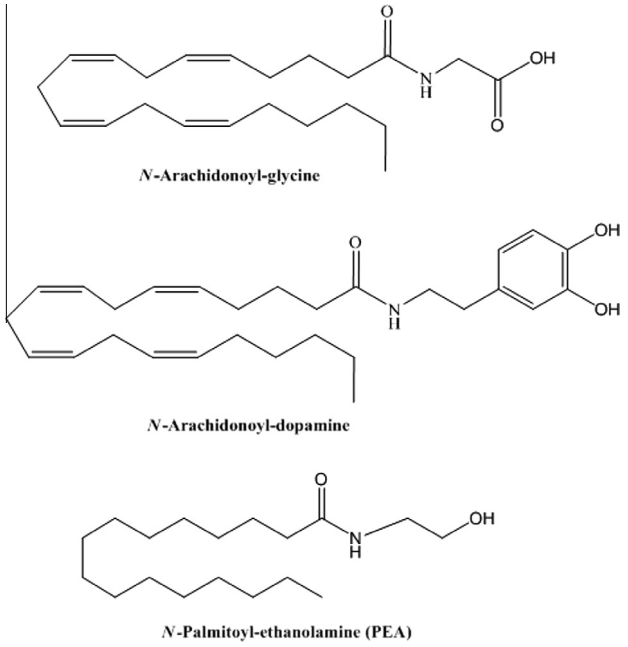


**Fig. 3.** Chemical structures of 3 among the most studied endocannabinoid-like mediators, *N*-arachidonoyl-glycine, *N*-arachidonoyl-dopamine, and *N*-palmitoyl-ethanolamine.

analgesic actions, seemingly mediated by $Ca^{2+}$-activated $K^+$ channels, and long-term (nuclear) antiinflammatory effects [48]; (2) potentiation of AEA actions at cannabinoid receptors and of AEA desensitization of TRPV1 channels ("entourage" effects), and possibly direct activation and desensitization of TRPV1 at higher concentrations [19]; (3) activation of the orphan GPCR, GPR55, or of PPAR-γ [7,14,68], although these mechanisms (and their involvement in the analgesic actions of PEA) are less established than the former 2; and 4) a combination of 2 or more of the above mechanisms [14]. While a full review of the several facets of PEA pharmacology in animal models of pain and inflammation was not among the aims of this article, it is important to underline that phase I and II trials have corroborated the efficacy of PEA and its formulations against: (1) entrapment neuropathy of the median nerve in the wrist [12]; (2) chronic pelvic pain related to endometriosis after laparoscopic assessment [11]; (3) temporomandibular joint inflammatory pain [51]; and (4) chronic pain severity in 610 patients, with pain associated to different pathological conditions [35]. Furthermore, enhancers of PEA endogenous levels through inhibition of its most important hydrolytic enzyme, the *N*-acylethanolamine-hydrolyzing acid amidase, are also being tested against pain and inflammation [69,72]. It is possible that, following the example of PEA, other multitarget ELMs might constitute in the future the bases to develop new analgesic treatments as "medical foods."

## 4. Back to cannabis: Phytocannabinoids

It seemed emblematic to end this review article by going back to where the whole story began, that is, to cannabis and cannabinoids. As mentioned above, studies on THC triggered the discovery of the "endocannabinoidome," but this compound is only one of the several cannabinoids (or "phytocannabinoids") so far identified in cannabis flowers (Fig. 1). The second most studied such compound is CBD, the antiinflammatory properties of which have been known since the end of last century and are now being investigated in their mechanistic aspects (see [42] for a recent review). Over the last 2 decades, in addition to its antioxidant properties, a plethora of molecular targets have been proposed for CBD, which could explain why this compound has proven efficacious in nearly all types of inflammatory conditions investigated thus far [42]. However, assuming that this could be a criterion for selection (given the fact that very high doses of CBD can be administered in vivo with little or no complication), only little more than a handful of these effects are exerted at sub/low-micromolar concentrations in vitro, including: (1) inhibition of the equilibrative nucleoside transporter and subsequent indirect activation of adenosine receptors [8]; (2) indirect activation of 5-$HT_{1a}$ receptors [62]; (3) inhibition of AEA cellular reuptake and hydrolysis by FAAH-1 [4,46] and subsequent indirect activation of cannabinoid receptors; (4) activation and desensitization of TRPV1 [4] as well as of TRPV type-2 [61] and transient receptor potential of ankyrin type-1 (TRPA1) channels [21]; (5) activation of PPAR-γ [29]; (6) inhibition of T-type $Ca^{2+}$ channels [65]; (7) activation of α3 glycine receptors [80]; and (8) antagonism of GPR55, at least according to some authors (see [71] for review). Moreover, only sparse evidence exists for the participation of these potential direct and indirect targets in the antiinflammatory actions of CBD, and such evidence is limited to TRPV1 [15,17], α3 glycine receptors [80], TRPA1, $CB_1$, 5-$HT_{1a}$, and adenosine A1 receptors [50].

More recently, other non-THC cannabinoids also have been the focus of pharmacological studies, and, in particular, for $\Delta^9$-tetrahydrocannabivarin (THCV), $\Delta^9$-tetrahydrocannabiorcol (THCO), cannabichromene (CBC), and cannabigerol (CBG) (Fig. 1), there starts to be evidence for analgesic and antiinflammatory actions. These phytocannabinoids all activate and desensitize TRPA1 channels, whereas only THCV and cannabigerol are known to also activate TRPV1 [1,21]. Furthermore, of these 4 compounds, only THCV was reported to bind to $CB_1$ and $CB_2$ receptors, although as a neutral antagonist rather than an agonist [76]. This property does not prevent THCV from exerting important antiinflammatory, analgesic, and antihyperalgesic actions in models of acute, neuropathic, and inflammatory pain (which, however, may be partly due also to THCV capability of activating $CB_2$ receptors at higher concentrations) [5]. Indeed, there is evidence that also $CB_1$ antagonists/inverse agonists produce paradoxical analgesic and antiinflammatory effects [16,18]. On the other hand, in a model of visceral pain, THCV was shown to antagonize THC analgesic effects [6]. Like CBD, CBC, injected locally, also stimulates the descending antinociceptive pathway in the periaqueductal grey of healthy mice via a combination of mechanisms including indirect or direct activation of TRPA1, $CB_1$, 5-$HT_{1a}$, and adenosine A1 receptors [50], and inhibits inflammation-induced hypermotility of the small intestine via unknown molecular mechanisms [43]. Unlike CBD, however, it inhibits visceral pain induced by acetic acid [6]. Spinally injected THCO, by activating TRPA1 and subsequently reducing voltage-gated calcium and sodium currents in primary sensory neurons, was reported to increase the paw withdrawal threshold in the paw-pressure test and paw withdrawal latency in the cold-plate test in mice [1]. Finally, preliminary evidence exists for antihyperalgesic and antioedemigenic action of cannabigerol in the carrageenan-induced model of inflammatory pain (Comelli et al., Symposium on Cannabinoids, Freiburg, Germany, July 22–27, 2012). These latter effects are mediated by α2-adrenoceptors, in agreement with the recently reported agonist effect of CBG at these receptors [9].

In summary, at least as far as their analgesic and antiinflammatory effects are concerned, non-THC cannabinoids seem to recapitulate the multi-target mechanism of action of AEA and ELMs. However, even with the most studied of these natural products (i.e., CBD), much work is still needed to fully understand what mechanisms are involved in what effects and, perhaps even more importantly, to ascertain whether such knowledge can be eventually translated into the development of novel analgesics. Nevertheless, perhaps emblematic of the need for a multitarget approach to tackle the as-yet unsolved problem of untreatable chronic and neuropathic pain conditions, the first phytocannabinoid-based drug, Sativex® (GW Pharmaceuticals, Salisbury, UK), has been approved not only

for the treatment of spasticity in multiple sclerosis in several countries, but, since 2007 in Canada, also against neuropathic pain in multiple sclerosis and cancer pain [63,67]. In fact, Sativex is a botanical drug constituted of an approximately 1:1 ratio of cannabinoid-enriched extracts from 2 cannabis varieties, one producing predominantly THC and the other CBD. It was initially developed based on data suggesting that the latter compound, in addition to its own therapeutic actions, also counteracts some of the psychotropic effects of THC, and hence allows for the administration of otherwise less tolerable dosages of this drug [67]. Thus, Sativex represents the first successful example, not only of a phytocannabinoid-based botanical drug, but also of a therapy exploiting the principle of the multitarget approach both to improve the efficacy and to reduce the side effects of a monotherapy. Indeed, when manipulating the endocannabinoid system, one should always give preference to approaches that produce as little change as possible in the overall activity of CB1 receptors in the brain, as indicated by the psychiatric side effects often observed with both CB1 receptor inverse agonists (depression and anxiety, in obese patients) and agonists (psychosis, especially in adolescents, and euphoria) [45].

## 5. Conclusions

We have tried in this article to show how, starting from studies on the medicinal properties of the cannabis plant, a whole system of endogenous chemical signals potentially involved in pain and inflammation has been revealed. The receptors and enzymes (e.g., orphan and already known GPCRs, TRP channels, PPARs, arachidonate cascade enzymes) that, together with these chemical signals, constitute the "endocannabinoidome" (Table 1), seem to act as molecular targets not only for THC but also for other previously neglected cannabinoids. The therapeutic use of THC and its synthetic analogue, nabilone, against pain has recently met with some success [74,77]. Safe as well as efficacious pharmacological interventions targeting this complex system, however, are difficult to develop, although not impossible, especially if one revives the idea of the multitarget drug, be it of synthetic or natural origin.

## Conflict of interest statement

The authors are recipients of research grants from GW Pharmaceuticals Ltd., UK. V.D. is a consultant for GW Pharmaceuticals Ltd., receives research funds from Allergan Plc., USA, and holds patents on the use of PEA or *N*-arachidonoyl-serotonin against pain.

## References


[1] Andersson DA, Gentry C, Alenmyr L, Killander D, Lewis SE, Andersson A, Bucher B, Galzi JL, Sterner O, Bevan S, Högestätt ED, Zygmunt PM. TRPA1 mediates spinal antinociception induced by acetaminophen and the cannabinoid Δ(9)-tetrahydrocannabiorcol. Nat Commun 2011;2:551.

[2] Bab I, Smoum R, Bradshaw H, Mechoulam R. Skeletal lipidomics: regulation of bone metabolism by fatty acid amide family. Br J Pharmacol 2011;163:1441–6.

[3] Barbara G, Alloui A, Nargeot J, Lory P, Eschalier A, Bourinet E, Chemin J. T-type calcium channel inhibition underlies the analgesic effects of the endogenous lipoamino acids. J Neurosci 2009;29:13106–14.

[4] Bisogno T, Hanus L, De Petrocellis L, Tchilibon S, Ponde DE, Brandi I, Moriello AS, Davis JB, Mechoulam R, Di Marzo V. Molecular targets for cannabidiol and its synthetic analogues: effect on vanilloid VR1 receptors and on the cellular uptake and enzymatic hydrolysis of anandamide. Br J Pharmacol 2001;134:845–52.

[5] Bolognini D, Costa B, Maione S, Comelli F, Marini P, Di Marzo V, Parolaro D, Ross RA, Gauson LA, Cascio MG, Pertwee RG. The plant cannabinoid Delta9-tetrahydrocannabivarin can decrease signs of inflammation and inflammatory pain in mice. Br J Pharmacol 2010;160:677–87.

[6] Booker L, Naidu PS, Razdan RK, Mahadevan A, Lichtman AH. Evaluation of prevalent phytocannabinoids in the acetic acid model of visceral nociception. Drug Alcohol Depend 2009;105:42–7.

[7] Cantarella G, Scollo M, Lempereur L, Saccani-Jotti G, Basile F, Bernardini R. Endocannabinoids inhibit release of nerve growth factor by inflammation-activated mast cells. Biochem Pharmacol 2011;82:380–8.

[8] Carrier EJ, Auchampach JA, Hillard CJ. Inhibition of an equilibrative nucleoside transporter by cannabidiol: a mechanism of cannabinoid immunosuppression. Proc Natl Acad Sci USA 2006;103:7895–900.

[9] Cascio MG, Gauson LA, Stevenson LA, Ross RA, Pertwee RG. Evidence that the plant cannabinoid cannabigerol is a highly potent alpha2-adrenoceptor agonist and moderately potent 5HT1A receptor antagonist. Br J Pharmacol 2010;159:129–41.

[10] Clifford DB, Simpson DM, Brown S, Moyle G, Brew BJ, Conway B, Tobias JK, Vanhove GF, NGX-4010 C119 Study Group. A randomized, double-blind, controlled study of NGX-4010, a capsaicin 8% dermal patch, for the treatment of painful HIV-associated distal sensory polyneuropathy. J Acquir Immune Defic Syndr 2012;59:126–33.

[11] Cobellis L, Castaldi MA, Giordano V, Trabucco E, De Franciscis P, Torella M, Colacurci N. Effectiveness of the association micronized *N*-Palmitoylethanolamine (PEA)-transpolydatin in the treatment of chronic pelvic pain related to endometriosis after laparoscopic assessment: a pilot study. Eur J Obstet Gynecol Reprod Biol 2011;158:82–6.

[12] Conigliaro R, Drago V, Foster PS, Schievano C, Di Marzo V. Use of palmitoylethanolamide in the entrapment neuropathy of the median in the wrist. Minerva Med 2011;102:141–7.

[13] Costa B, Bettoni I, Petrosino S, Comelli F, Giagnoni G, Di Marzo V. The dual fatty acid amide hydrolase/TRPV1 blocker, *N*-arachidonoyl-serotonin, relieves carrageenan-induced inflammation and hyperalgesia in mice. Pharmacol Res 2010;61:537–46.

[14] Costa B, Comelli F, Bettoni I, Colleoni M, Giagnoni G. The endogenous fatty acid amide, palmitoylethanolamide, has anti-allodynic and anti-hyperalgesic effects in a murine model of neuropathic pain: involvement of CB(1), TRPV1 and PPARgamma receptors and neurotrophic factors. PAIN® 2008;139:541–50.

[15] Costa B, Giagnoni G, Franke C, Trovato AE, Colleoni M. Vanilloid TRPV1 receptor mediates the antihyperalgesic effect of the nonpsychoactive cannabinoid, cannabidiol, in a rat model of acute inflammation. Br J Pharmacol 2004;143:247–50.

[16] Costa B, Trovato AE, Colleoni M, Giagnoni G, Zarini E, Croci T. Effect of the cannabinoid CB1 receptor antagonist, SR141716, on nociceptive response and nerve demyelination in rodents with chronic constriction injury of the sciatic nerve. PAIN® 2005;116:52–61.

[17] Costa B, Trovato AE, Comelli F, Giagnoni G, Colleoni M. The non-psychoactive cannabis constituent cannabidiol is an orally effective therapeutic agent in rat chronic inflammatory and neuropathic pain. Eur J Pharmacol 2007;556:75–83.

[18] Croci T, Zarini E. Effect of the cannabinoid CB1 receptor antagonist rimonabant on nociceptive responses and adjuvant-induced arthritis in obese and lean rats. Br J Pharmacol 2007;150:559–66.

[19] De Petrocellis L, Davis JB, Di Marzo V. Palmitoylethanolamide enhances anandamide stimulation of human vanilloid VR1 receptors. FEBS Lett 2001;506:253–6.

[20] De Petrocellis L, Di Marzo V. Non-CB1, non-CB2 receptors for endocannabinoids, plant cannabinoids, and synthetic cannabimimetics: focus on G-protein-coupled receptors and transient receptor potential channels. J Neuroimmune Pharmacol 2010;5:103–21.

[21] De Petrocellis L, Ligresti A, Moriello AS, Allarà M, Bisogno T, Petrosino S, Stott CG, Di Marzo V. Effects of cannabinoids and cannabinoid-enriched Cannabis extracts on TRP channels and endocannabinoid metabolic enzymes. Br J Pharmacol 2011;163:1479–94.

[22] Devane WA, Hanus L, Breuer A, Pertwee RG, Stevenson LA, Griffin G, Gibson D, Mandelbaum A, Etinger A, Mechoulam R. Isolation and structure of a brain constituent that binds to the cannabinoid receptor. Science 1992;258:1946–9.

[23] Di Marzo V. Endocannabinoids: synthesis and degradation. Rev Physiol Biochem Pharmacol 2008;160:1–24.

[24] Di Marzo V. Inhibitors of endocannabinoid breakdown for pain: not so FA(AH)cile, after all. PAIN® 2012;53:1785–6.

[25] Di Marzo V. Targeting the endocannabinoid system: to enhance or reduce? Nat Rev Drug Discov 2008;7:438–55.

[26] Di Marzo V, Fontana A. Anandamide, an endogenous cannabinomimetic eicosanoid: 'killing two birds with one stone'. Prostaglandins Leukot Essent Fatty Acids 1995;53:1–11.

[27] Edington AR, McKinzie AA, Reynolds AJ, Kassiou M, Ryan RM, Vandenberg RJ. Extracellular loops 2 and 4 of GLYT2 are required for *N*-arachidonylglycine inhibition of glycine transport. J Biol Chem 2009;284:36424–30.

[28] Esposito E, Cuzzocrea S. Palmitoylethanolamide is a new possible pharmacological treatment for the inflammation associated with trauma. Mini Rev Med Chem 2013;13:237–55.

[29] Esposito G, Scuderi C, Valenza M, Togna GI, Latina V, De Filippis D, Cipriano M, Carratù MR, Iuvone T, Steardo L. Cannabidiol reduces Aβ-induced neuroinflammation and promotes hippocampal neurogenesis through PPARγ involvement. PLoS One 2011;6:e28668.

[30] Facci L, Dal Toso R, Romanello S, Buriani A, Skaper SD, Leon A. Mast cells express a peripheral cannabinoid receptor with differential sensitivity to anandamide and palmitoylethanolamide. Proc Natl Acad Sci USA 1995;92:3376–80.

[31] Farkas I, Tuboly G, Benedek G, Horvath G. The antinociceptive potency of *N*-arachidonoyl-dopamine (NADA) and its interaction with endomorphin-1 at the spinal level. Pharmacol Biochem Behav 2011;99:731–7.

[32] Feledziak M, Lambert DM, Marchand-Brynaert J, Muccioli GG. Inhibitors of the endocannabinoid-degrading enzymes, or how to increase endocannabinoid's activity by preventing their hydrolysis. Recent Pat CNS Drug Discov 2012;7:49–70.

[33] Gaoni Y, Mechoulam R. Isolation, structure and partial synthesis of an active constituent of hashish. J Am Chem Soc 1964;86:1646–7.
[34] Gatta L, Piscitelli F, Giordano C, Boccella S, Lichtman A, Maione S, Di Marzo V. Discovery of prostamide F2α and its role in inflammatory pain and dorsal horn nociceptive neuron hyperexcitability. PLoS One 2012;7:e31111.
[35] Gatti A, Lazzari M, Gianfelice V, Di Paolo A, Sabato E, Sabato AF. Palmitoylethanolamide in the treatment of chronic pain caused by different etiopathogenesis. Pain Med 2012;13:1121–30.
[36] Gilmore AJ, Heblinski M, Reynolds A, Kassiou M, Connor M. Inhibition of human recombinant T-type calcium channels by *N*-arachidonoyl serotonin. Br J Pharmacol 2012;167:1076–88.
[37] Giordano C, Cristino L, Luongo L, Siniscalco D, Petrosino S, Piscitelli F, Marabese I, Gatta L, Rossi F, Imperatore R, Palazzo E, de Novellis V, Di Marzo V, Maione S. TRPV1-dependent and -independent alterations in the limbic cortex of neuropathic mice: impact on glial caspases and pain perception. Cereb Cortex 2012;22:2495–518.
[38] Glaser ST, Kaczocha M. Cyclooxygenase-2 mediates anandamide metabolism in the mouse brain. J Pharmacol Exp Ther 2010;335:380–8.
[39] Huang SM, Bisogno T, Petros TJ, Chang SY, Zavitsanos PA, Zipkin RE, Sivakumar R, Coop A, Maeda DY, De Petrocellis L, Burstein S, Di Marzo V, Walker JM. Identification of a new class of molecules, the arachidonyl amino acids, and characterization of one member that inhibits pain. J Biol Chem 2001;276:42639–44.
[40] Huang SM, Bisogno T, Trevisani M, Al-Hayani A, De Petrocellis L, Fezza F, Tognetto M, Petros TJ, Krey JF, Chu CJ, Miller JD, Davies SN, Geppetti P, Walker JM, Di Marzo V. An endogenous capsaicin-like substance with high potency at recombinant and native vanilloid VR1 receptors. Proc Natl Acad Sci USA 2002;99:8400–5.
[41] Huggins JP, Smart TS, Langman S, Taylor L, Young T. An efficient randomised, placebo-controlled clinical trial with the irreversible fatty acid amide hydrolase-1 inhibitor PF-04457845, which modulates endocannabinoids but fails to induce effective analgesia in patients with pain due to osteoarthritis of the knee. PAIN® 2012;153:1837–46.
[42] Izzo AA, Borrelli F, Capasso R, Di Marzo V, Mechoulam R. Non-psychotropic plant cannabinoids: new therapeutic opportunities from an ancient herb. Trends Pharmacol Sci 2009;30:515–27.
[43] Izzo AA, Capasso R, Aviello G, Borrelli F, Romano B, Piscitelli F, Gallo L, Capasso F, Orlando P, Di Marzo V. Inhibitory effect of cannabichromene, a major non-psychotropic cannabinoid extracted from Cannabis sativa, on inflammation-induced hypermotility in mice. Br J Pharmacol 2012;166:1444–60.
[44] Kohno M, Hasegawa H, Inoue A, Muraoka M, Miyazaki T, Oka K, Yasukawa M. Identification of *N*-arachidonylglycine as the endogenous ligand for orphan G-protein-coupled receptor GPR18. Biochem Biophys Res Commun 2006;347:827–32.
[45] Kunos G, Osei-Hyiaman D, Bátkai S, Sharkey KA, Makriyannis A. Should peripheral CB(1) cannabinoid receptors be selectively targeted for therapeutic gain? Trends Pharmacol Sci 2009;30:1–7.
[46] Leweke FM, Piomelli D, Pahlisch F, Muhl D, Gerth CW, Hoyer C, Klosterkötter J, Hellmich M, Koethe D. Cannabidiol enhances anandamide signaling and alleviates psychotic symptoms of schizophrenia. Transl Psychiatry 2012;2:e94.
[47] Ligresti A, Petrosino S, Di Marzo V. From endocannabinoid profiling to 'endocannabinoid therapeutics'. Curr Opin Chem Biol 2009;13:321–31.
[48] LoVerme J, Russo R, La Rana G, Fu J, Farthing J, Mattace-Raso G, Meli R, Hohmann A, Calignano A, Piomelli D. Rapid broad-spectrum analgesia through activation of peroxisome proliferator-activated receptor-alpha. J Pharmacol Exp Ther 2006;319:1051–61.
[49] Maione S, De Petrocellis L, de Novellis V, Moriello AS, Petrosino S, Palazzo E, Rossi FS, Woodward DF, Di Marzo V. Analgesic actions of *N*-arachidonoyl-serotonin, a fatty acid amide hydrolase inhibitor with antagonistic activity at vanilloid TRPV1 receptors. Br J Pharmacol 2007;150:766–81.
[50] Maione S, Piscitelli F, Gatta L, Vita D, De Petrocellis L, Palazzo E, de Novellis V, Di Marzo V. Non-psychoactive cannabinoids modulate the descending pathway of antinociception in anaesthetized rats through several mechanisms of action. Br J Pharmacol 2011;162:584–96.
[51] Marini I, Bartolucci ML, Bortolotti F, Gatto MR, Bonetti GA. Palmitoylethanolamide versus a nonsteroidal anti-inflammatory drug in the treatment of temporomandibular joint inflammatory pain. J Orofac Pain 2012;26:99–104.
[52] Matsuda LA, Lolait SJ, Brownstein MJ, Young AC, Bonner TI. Structure of a cannabinoid receptor and functional expression of the cloned cDNA. Nature 1990;346:561–4.
[53] Mechoulam R, Ben-Shabat S, Hanus L, Ligumsky M, Kaminski NE, Schatz AR, Gopher A, Almog S, Martin BR, Compton DR, Pertwee RG, Griffin G, Bayewitch M, Barg J, Vogel Z. Identification of an endogenous 2-monoglyceride, present in canine gut, that binds to cannabinoid receptors. Biochem Pharmacol 1995;50:83–90.
[54] Mechoulam R, Shvo Y. Hashish. I. The structure of cannabidiol. Tetrahedron 1963;19:2073–8.
[55] Micale V, Cristino L, Tamburella A, Petrosino S, Leggio GM, Drago F, Di Marzo V. Anxiolytic effects in mice of a dual blocker of fatty acid amide hydrolase and transient receptor potential vanilloid type-1 channels. Neuropsychopharmacology 2009;34:593–606.
[56] Morera E, De Petrocellis L, Morera L, Moriello AS, Ligresti A, Nalli M, Woodward DF, Di Marzo V, Ortar G. Synthesis and biological evaluation of piperazinyl carbamates and ureas as fatty acid amide hydrolase (FAAH) and transient receptor potential (TRP) channel dual ligands. Bioorg Med Chem Lett 2009;19:6806–9.
[57] Munro S, Thomas KL, Abu-Shaar M. Molecular characterization of a peripheral receptor for cannabinoids. Nature 1993;365:61–5.
[58] Nomura DK, Morrison BE, Blankman JL, Long JZ, Kinsey SG, Marcondes MC, Ward AM, Hahn YK, Lichtman AH, Conti B, Cravatt BF. Endocannabinoid hydrolysis generates brain prostaglandins that promote neuroinflammation. Science 2011;334:809–13.
[59] O'Shaughnessy WB. On the preparations of the Indian hemp or gunjah (*Cannabis indica*); their effects on the animal system in health, and their utility in the treatment of tetanus and other convulsive diseases. Trans Med Phys Soc Bengal 1838–1840;71–102:421–61.
[60] Piscitelli F, Di Marzo V. "Redundancy" of endocannabinoid inactivation: new challenges and opportunities for pain control. ACS Chem Neurosci 2012;3:356–63.
[61] Qin N, Neeper MP, Liu Y, Hutchinson TL, Lubin ML, Flores CM. TRPV2 is activated by cannabidiol and mediates CGRP release in cultured rat dorsal root ganglion neurons. J Neurosci 2008;28:6231–8.
[62] Rock EM, Bolognini D, Limebeer CL, Cascio MG, Anavi-Goffer S, Fletcher PJ, Mechoulam R, Pertwee RG, Parker LA. Cannabidiol, a non-psychotropic component of cannabis, attenuates vomiting and nausea-like behaviour via indirect agonism of 5-HT(1A) somatodendritic autoreceptors in the dorsal raphe nucleus. Br J Pharmacol 2012;165:2620–34.
[63] Rog DJ, Nurmikko TJ, Young CA. Oromucosal delta9-tetrahydrocannabinol/cannabidiol for neuropathic pain associated with multiple sclerosis: an uncontrolled, open-label, 2-year extension trial. Clin Ther 2007;29:2068–79.
[64] Ross HR, Gilmore AJ, Connor M. Inhibition of human recombinant T-type calcium channels by the endocannabinoid *N*-arachidonoyl dopamine. Br J Pharmacol 2009;156:740–50.
[65] Ross HR, Napier I, Connor M. Inhibition of recombinant human T-type calcium channels by Delta9-tetrahydrocannabinol and cannabidiol. J Biol Chem 2008;283:16124–34.
[66] Rouzer CA, Marnett LJ. Non-redundant functions of cyclooxygenases: oxygenation of endocannabinoids. J Biol Chem 2008;283:8065–9.
[67] Russo EB, Guy GW, Robson PJ. Cannabis, pain, and sleep: lessons from therapeutic clinical trials of Sativex, a cannabis-based medicine. Chem Biodivers 2007;4:1729–43.
[68] Ryberg E, Larsson N, Sjögren S, Hjorth S, Hermansson NO, Leonova J, Elebring T, Nilsson K, Drmota T, Greasley PJ. The orphan receptor GPR55 is a novel cannabinoid receptor. Br J Pharmacol 2007;152:1092–101.
[69] Saturnino C, Petrosino S, Ligresti A, Palladino C, De Martino G, Bisogno T, Di Marzo V. Synthesis and biological evaluation of new potential inhibitors of *N*-acylethanolamine hydrolyzing acid amidase. Bioorg Med Chem Lett 2010;20:1210–3.
[70] Schlosburg JE, Blankman JL, Long JZ, Nomura DK, Pan B, Kinsey SG, Nguyen PT, Ramesh D, Booker L, Burston JJ, Thomas EA, Selley DE, Sim-Selley LJ, Liu QS, Lichtman AH, Cravatt BF. Chronic monoacylglycerol lipase blockade causes functional antagonism of the endocannabinoid system. Nat Neurosci 2010;13:1113–9.
[71] Sharir H, Abood ME. Pharmacological characterization of GPR55, a putative cannabinoid receptor. Pharmacol Ther 2010;126:301–13.
[72] Solorzano C, Zhu C, Battista N, Astarita G, Lodola A, Rivara S, Mor M, Russo R, Maccarrone M, Antonietti F, Duranti A, Tontini A, Cuzzocrea S, Tarzia G, Piomelli D. Selective *N*-acylethanolamine-hydrolyzing acid amidase inhibition reveals a key role for endogenous palmitoylethanolamide in inflammation. Proc Natl Acad Sci U S A 2009;106:20966–71.
[73] Sugiura T, Kondo S, Sukagawa A, Nakane S, Shinoda A, Itoh K, Yamashita A, Waku K. 2-Arachidonoylglycerol: a possible endogenous cannabinoid receptor ligand in brain. Biochem Biophys Res Commun 1995;215:89–97.
[74] Svendsen KB, Jensen TS, Bach FW. Effect of the synthetic cannabinoid dronabinol on central pain in patients with multiple sclerosis. Ugeskr Laeger 2005;167:2772–4.
[75] Szallasi A, Sheta M. Targeting TRPV1 for pain relief: limits, losers and laurels. Expert Opin Investig Drugs 2012;21:1351–69.
[76] Thomas A, Stevenson LA, Wease KN, Price MR, Baillie G, Ross RA, Pertwee RG. Evidence that the plant cannabinoid Delta9-tetrahydrocannabivarin is a cannabinoid CB1 and CB2 receptor antagonist. Br J Pharmacol 2005;146:917–26.
[77] Toth C, Mawani S, Brady S, Chan C, Liu C, Mehina E, Garven A, Bestard J, Korngut L. An enriched-enrolment, randomized withdrawal, flexible-dose, double-blind, placebo-controlled, parallel assignment efficacy study of nabilone as adjuvant in the treatment of diabetic peripheral neuropathic pain. PAIN® 2012;153:2073–82.
[78] Verhoeckx KC, Voortman T, Balvers MG, Hendriks HF, Wortelboer HM, Witkamp RF. Presence, formation and putative biological activities of *N*-acyl serotonins, a novel class of fatty-acid derived mediators, in the intestinal tract. Biochim Biophys Acta 2011;1811:578–86.
[79] Vuong LA, Mitchell VA, Vaughan CW. Actions of *N*-arachidonyl-glycine in a rat neuropathic pain model. Neuropharmacology 2008;54:189–93.
[80] Xiong W, Cui T, Cheng K, Yang F, Chen SR, Willenbring D, Guan Y, Pan HL, Ren K, Xu Y, Zhang L. Cannabinoids suppress inflammatory and neuropathic pain by targeting α3 glycine receptors. J Exp Med 2012;209:1121–34.